(b) contacting the cells with mitogenic antibodies to induce cell activation;

(c) selecting CD4+ cells that are HIV- after activation; and

(d) inducing cell proliferation and expanding the selected cells to in excess of 1×10^{10} cells per liter, wherein:

cell proliferation and expansion is performed in the absence of exogenous interleukin-2 (IL-2), wherein, in the contacting step, the activation of the cells occurs under conditions that promote Th1 cell differentiation to produce a population of cells that contains predominantly Th1 cells.

38. (Three Times Amended) The method of claim 37, further comprising:

after selecting CD4+ cells that are HIV- and prior to expanding the selected cells, growing a plurality of aliquots in the presence of mitogenic agents;

selecting from the aliquots those that are HIV-; and

then expanding the selected cells to in excess of 1×10^{10} cells per liter.

39. (Twice Amended) The method of claim 37, wherein the cells are activated with anti-CD3 monoclonal antibodies in the presence of interferon-γ (IFN-γ).

154. (Amended) The method of claim 40, wherein cell expansion is effected in a hollow fiber bioreactor.

REMARKS

A check for the fee for a three month extension of time accompanies this response. Any fee that may be due in connection with this application may be charged to Deposit Account No. Deposit Account No. 50-1213. If a Petition for extension of time is needed, this paper is to be considered such Petition.

A Change of Address of the undersigned has been filed under separate cover.

Claims 36-40 and 154-167 are presently pending. Claims 36, 38, 40, 154-157, 158, 162-164 and 166 have been previosly withdrawn from condideration.

Claims 36, 155, 156, 157, 158, 159 and 162 are cancelled herein without prejudice as being withdrawn from consideration as allegedly drawn to non-elected subject matter. However, previously withdrawn claims 38, 40, 154, 163, 164 and 166 are now dependent upon previously elected claims 37 and 39, and thus remain presently pending. Applicant reserves the right to file divisional applications to the withdrawn subject matter; the Office is reminded that as between any of the cancelled claims and the presently pending claims obviousness-type double patenting cannot be held. Applicant reserves the right to file divisional applications to any cancelled subject matter.

The amendment to claims 37 and 38 to reflect in excess of 10^{10} cells/liter finds basis in previous claims 155, 159 and 162, which have been cancelled herein. A marked up copy of claims showing the amendments herein is appended hereto. Entry of the amendments is respectfully submitted to be proper because the amendments are believed to place the claims in condition for allowance, or in the alternative, reduce the number of issues on appeal. A Notice of Appeal is filed herewith.

THE REJECTION OF CLAIMS 37, 39, 159-161, 165 and 167 UNDER 35 U.S.C. §112, FIRST PARAGRAPH

The rejection of claims 37, 39, 159-161, 165 and 167 under 35 U.S.C. 112, first paragraph, as allegedly containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled are that the inventor, at the time the application was file, had possession of the claimed invention; is respectfully traversed. It is respectfully submitted that this rejection has been rendered moot by the amendments to claims 37 and 39 herewith, whereby the term "isolating" has been replaced with the term "collecting," which are believed to have the same meaning and scope within the context of the claimed methods. It is respectfully submitted that the term "collecting," in the context of collecting mononuclear cells, finds support in

Applicant's specification, at page 28, line 20 through page 29, line 5, which states:

> **C. Methods for production of regulatory cells**
> A method for obtaining regulatory cells for use in ACT protocols is provided herein. A method for obtaining effector cells for use in ACT protocols without the need for exogenous agents, such as IL-2, that sustain the viability of such cells is also provided. The method includes some or all of the following steps: (1) **collecting mononuclear cells** from a patient; (2) treating the cells ex vivo with that agents that cause some or all of the cells to the differentiate into desired T cell subtypes; (3) purifying the resulting cells; and (4) expanding these cells by contacting them with a mitogenic agent that specifically interacts with a cell surface receptor. Such agents are herein preferably mitogenic monoclonal antibodies. The expanded cells may be further purified to select for the desired subtype.
> 1. ***Collecting*** **mononuclear cells**
> Mononuclear cells (i.e., lymphocytes and monocytes) can be obtained from a variety of sources, including, but not limited to,

See also, Applicant's specification, at Example 3, section A., which states:

> **A. Obtaining Mononuclear Cells**
> An HIV^+ patient, identified by a routine blood screening procedure confirmed by Western Blot analysis, in WHO stage IV was the donor for this study. The patient underwent a leukopheresis procedure for **collection** of peripheral blood mononuclear cells.

Accordingly, in view of the amendments to claims 37 and 39, reconsideration and withdrawal of this rejection is respectfully requested.

THE REJECTION OF CLAIMS 37, 39, 159-161, 165 and 167 UNDER 35 U.S.C. §112, FIRST PARAGRAPH

The rejection of claims 37, 39, 159-161, 165 and 167 under 35 U.S.C. 112, first paragraph, as allegedly containing subject matter which was not described in the specification in such a way as to reasonably convey to one skilled are that the inventor, at the time the application was file, had possession of the

claimed invention, is respectfully traversed. The Examiner urges that there is no "support in the specification for recitation of "mitogenic antibodies" in claims 36 and 37.

It is respectfully submitted that there is indeed basis in this application and in the parent application for the phrase "mitogenic antibodies." Attention is directed, for example, to Applicant's specification, at page 18, lines 10-16, which states:

> Suitable **mitogenic antibodies** may be identified empirically by testing selected antibodies singly or in combination for the ability to increase numbers of a specific effector cell. Suitable **mitogenic antibodies** or combinations thereof will increase the number of cells in a selected time period, typically 1 to 10 days, by at least about 50%, preferably about 100% and more preferably 150-200% or more, compared to the numbers of cells in the absence of the antibody.

Thus, the Examiner's assertion regarding written description, at p. 3, lines 11-12 of the Official Action, that "[t]here is no support in the specification for the recitation of 'mitogenic antibodies' in claim 37" **is not correct.** Accordingly, reconsideration and withdrawal of this rejection is respectfully requested.

THE REJECTION OF CLAIMS 37 AND 39 UNDER 35 U.S.C. §112, SECOND PARAGRAPH

Claims 37 and 39 are rejected under 35 U.S.C. 112, second paragraph, as being dependent on claim 36, which the Examiner alleges is "non-elected." This rejection has been rendered moot by the amendments to claims 37 and 39 herewith, such that they no longer depend from claim 36. Accordingly, reconsideration and withdrawal of this rejection is respectfully requested.

THE PRIORITY DATE

Regarding the priority date for the claimed invention, although Applicant maintains that the instantly claimed subject matter finds basis in the parent application, because none of the cited prior art refences have effective publication dates that are intervening between grandparent application USSN 08/506,668 (filed July 26, 1995) and parent application USSN 08/700,565 (filed July 25, 1996), the issue related to the priority date of the pending claims is moot.

REJECTION OF CLAIMS 37, 39 and 159-161 UNDER 35 U.S.C. § 103(a)

Claims 37, 39 and 159-161 are rejected under 35 U.S.C. § 103(a) as allegedly unpatentable over O'garra *et al.*, 1994, Current Opinion in Immun., 6:458-466 in view June *et al.* (WO 94/29436) or June *et al.* (USP 5,858,358) and Carew (USP 5,123,901) and Nabel *et al.*, 1987, Nature, 326(6114):711-71,is respectfully traversed.

The Claims

Claim 37 is directed to a method of producing virally purged CD4+ cells by isolating mononuclear cells from a patient infected with HIV, activating the cells by contacting the cells with mitogenic antibodies, selecting CD4+ cells that are HIV- after activation, and then expanding the selected cells to clinically relevant numbers in excess of 10^{10} in the absence of interleukin-2 (IL-2), wherein the cells are activated under conditions that produce Th1 cells. Claim 38 now depends on 37 and is directed to the method where after the selecting step and prior to expanding the selected cells, a plurality of aliquots of the cells are grown in the presence of mitogenic agents, and HIV- cells are selected and then expanded. Claims 39 and 40 specifies the reagents used to activate and grow the cells. Claims 154 and 165-167 recite that the cells are expanded in a hollow fiber bioreactor, and claims 160-161 and 163 and 164 recite the resulting volume and/or density.

Teachings of the cited references and differences from the instant claims

O'Garra *et al.*

O'Garra *et al.* is directed to a study to assess the role of cytokines in determining T-lymphocyte function. O'Garra *et al.* teaches that encounter with a host antigen can result in either cell-mediated or humoral classes of immune response and that these responses are attributable to the heterogeneity of $CD4^+$ T cells. O'Garra *et al.* further teaches that mouse $CD4^+$ T cell clones can be divided into two predominant cytokine secretion profiles designated Th1, which produce IL-2 and IFN-γ and other factors that promote delayed-type hypersensitivity reactions, and Th2, which produce Il-4, Il-5 and IL-10. The subsets by virtue of the differing cytokine profiles cross-regulate immune responses. O'Garra *et al.* merely speculates (page 459, col. 1) that the ability to control the emerging Th cell phenotype [*in vivo*] following exposure to antigen offers the potential to induce a response appropriate for each pathogen. O'Garra presents the results of studies designed to elucidate the pathways by which each type of subset is induced. O'Garra *et al.* concludes (page 462):

> . . . The question of whether Th1 and Th2 cells all arise from a common precursor, possibly a Th0-type cell, and whether such populations are malleable or can be differentiated further, remains ***an unresolved issue***, with important implications for the treatment of chronic disease. (emphasis added)

Thus, O'Garra *et al.* certainly does not teach any conditions that promote Th1 cell differentiation to produce a population of cells that contains predominantly Th1 cells, as required by the instant claims. Moreover, as acknowledged by the Examiner at page 5, lines 7-8 of the Office Action, "O'Garra *et al.* do not teach the claimes method of producing virally purged Th1 cells with the numbers of cells recited in the claims."

Therefore, because O'Garra *et al.* concludes that it is not clear whether Th1 and Th2 phenotypes can be altered, it certainly does not teach or suggest production of compositions of predominantly Th1, nor does O'Garra *et al.* provide

any motivation to produce large numbers or high densities of "virally purged" cells of any type.

It is respectfully submitted that neither of the secondary references, taken alone or in combination, cures the deficiencies in the teachings of O'garra. Therefore, the Examiner has failed to set forth a *prima facie* case of obviousness.

June *et al.* (WO 94/29436 or USP 5,858,358)

These two references are cumulative and are addressed together. The Examiner alleges, at page 5, lines 8-12 of the Official Action, that:

> June *et al.* (WO 94/29436)(Figures 1 and 2, pages 4-35) or June *et al.* (US Patent 5,858,358)(Figures 1 and 2, columns 4-32) both teach methods of expanding T cells to clinically relevant numbers without using exogenous growth factors (see abstract). Figures 1 and 2 of said publications show expansion of T cells to greater than 10^{10} cells.

However, June *et al.* does not teach growth of T cells to high cell density exceeding 10^{10} cells/Liter to obtain clinically relevant numbers of cells, as required by Applicants' claims. The data shown in Figures 1-3 of June *et al.* are merely extrapolations of overall growth from a single flask after bi-daily dilutions of the cells. For example, the reference teaches that the cells were maintained at "a cell density of 0.5×10^6/ml" (see, e.g., WO 94/29436 page 27, lns. 7-9 generally; page 27, lns. 35-36 regarding Figure 1; page 28, lns. 30-32 regarding Figure 2; and page 29, lns. 13-18 regarding Figure 3). Nowhere, does June *et al.* teach or suggest any cell densities in excess of 10^{10} per liter. Thus, June *et al.* does not teach or suggest growth of T cells under conditions that produce high cell densities in excess of 10^{10} cells/liter, to obtain clinically relevant numbers of cells, as required by Applicant's claims. June *et al.* merely discloses that its method can be used to expand T cells in long term tissue culture to obtain a population **increased in number** from about 100 to about 100,000 fold over the original starting cell population, **not increased in cell density**. Thus, Figures 1-3 actually

only show that the method results in cell numbers that could exceed 1 x 10^{10} cells, but in no way teaches or suggests preparation of such cells at a density of about 10^{10}/liter or more, as required by applicants' claims.

Moreover, the teachings of the June *et al.* references, e.g., at page 9 first complete paragraph of June *et al.* '29436, does not teach differentiation of T cells into Th1 cells prior to expansion. In this paragraph, June *et al.* describes antigen specific activation of a population of cells. Preparing antigen-specific cells does not involve differentiation of cells to produce Th1 cells as claimed in the instant claims. Antigen-specific activation of a population of cells is not the same as production of Th1 cells. Thus, June *et al.*, alone or in combination with O'Garra *et al.*, Nabel and Carew, does not teach or suggest any conditions that promote Th1 cell differentiation to produce a population of cells that contains predominantly Th1 cells, as required by the instant claims.

Carew

Carew does not cure the deficiencies in O'garra *et al.* and June *et al.* Carew is directed to a method for removing pathogenic agents from body fluids. The body fluid is perfused into a mixing coil with paramagnetic beads that selectively bind to the pathogenic agent; and the beads are then separated from the fluid. In one embodiment blood is treated to remove infected T-lymphocytes by continuously perfusing the blood through the mixing coil using a peristaltic pump. Thus, the method of Carew involves filtration of the blood of an individual.

Carew does not teach or suggest use of its method in combination with adoptive immunotherapy protocols in which selected cells are first activated and differentiated, and then expanded. Nor does Carew teach or suggest a step of expanding the Th1 cells to in excess of 10^{10} cells/liter in the absence of IL-2. Thus, Carew does not cure the deficiencies in the teachings of O'garra *et al.* and June *et al.*

Nabel *et al.*

Nabel *et al.* does not cure the deficiencies in the teachings of O'garra *et al.*, June *et al.* and Carew. Nabel *et al.* merely teaches that human immunodeficiency virus (HIV) production from latently infected T lymphocytes can be induced with compounds that activate the cells to secrete lymphokines. Because Nabel *et al.* provides no teachings or suggestions directed to methods for preparing HIV-compositions of cells, Nabel *et al.* has no relevance to the instant claims, which are directed to methods for preparing compositions of virally purged cells.

The Examiner has failed to set forth a case of *prima facie* obviousness

(1) Relevant law

In order to set forth a prima facie case of obviousness under 35 U.S.C. § 103: (1) there must be some teaching, suggestion or incentive supporting the combination of cited references to produce the claimed invention (ACS Hospital Systems, Inc. v. Montefiore Hospital, 732 F.2d 1572, 1577, 221 USPQ 329, 933 (Fed. Cir. 1984)) and (2) the combination of the cited references must actually teach or suggest the claimed invention. Further, that which is within the capabilities of one skilled in the art is not synonymous with that which is obvious. Ex parte Gerlach, 212 USPQ 471 (BPAI 1980). Obviousness is tested by "what the combined teachings of the references would have suggested to those of ordinary skill in the art" In re Keller, 642 F.2d 413, 425, 208 USPQ 871, 881 (CCPA 1981), but it cannot be established by combining the teachings of the prior art to produce the claimed invention, absent some teaching or suggestion supporting the combination (ACS Hosp. Systems, Inc. v Montefiore Hosp. 732 F.2d 1572, 1577. 221 USPQ 329, 933 (Fed. Cir. 1984)).

There would have been no motivation to have combined the teachings of O'garra *et al.* with June *et al.*, Carew and Nable, and the combination does not result in the instantly claimed methods

Motivation

None of the cited references, taken alone or in any combination thereof, teaches or suggests anything regarding preparation of **populations of cells that contain predominantly Th1 cells**. None of the cited references suggests any method that involves restoration of the immune system balance by administration of compositions containing substantially only one type of regulatory immune cell corresponding to Th1. It is respectfully submitted that only the instant application provides such suggestion and motivation.

O'Garra *et al.* describe properties of Th1 and Th2 cells *in vivo* and explains *in vivo* establishment of lymphokine-producing phenotypes and development thereof. As set forth above, O'Garra *et al.* certainly does not teach any conditions that promote Th1 cell differentiation to produce a population of cells that contains predominantly Th1 cells, as required by the instant claims.

The secondary references: June *et al.* is directed to methods for expansion of cells, but does not suggest expansion of one type of cell for any purpose, nor the expansion of cells to a density in excess of 10^{10} cells/liter. As discussed above, the cells produced by the method of June *et al.* are clearly not predominantly Th1 cells. The cytokine profile of the resulting cells (Table 2) is clearly that of a mixed population. Carew is directed to a perfusion-based method of removing pathogens from body fluids, and Nabel *et al.* teaches that compounds that activate lymphocytes can induce production of HIV from latently infected cells. Thus, Nabel *et al.* is merely directed to an observation.

Moreover, none of the art of record provides any uses for expanded compositions of predominantly Th1 cells. Thus, there is nothing in the teachings of the secondary references and O'garra *et al.* that would have motivated combination of their teachings nor that would have motivated one of ordinary skill to produce a predominantly Th1 cell population in excess of 10^{10} cells/liter.

The combination of references does not result in the claimed subject matter

Notwithstanding the lack of motivation, even if combined, the combination of cited references do not teach, suggest or result in differentiation of mononuclear cells to predominantly Th1 cells and the expansion of Th1 cells *in vitro* to produce compositions containing clinically relevant numbers of Th1 cells in excess of 10^{10} cells/liter, as required by Applicant's claims. Moreover, with respect to dependent claims, the combination of references do not disclose or suggest a density of more than 10^8 cells/ml of predominantly Th1 cells. Therefore, the Examiner has failed to set forth a prima facie case of obviousness.

The Rejection over O'garra *et al.* in view of June *et al.*, Carew and Nable *et al.* is Based on Improper Use of Hindsight.

The disclosure of the applicant cannot be used to hunt through the prior art for the claimed elements and then combine them as claimed. In re Laskowski, 871 F.2d 115, 117, 10 USPQ2d 1397, 1398 (Fed. Cir. 1989). "To imbue one of ordinary skill in the art with knowledge of the invention in suit, when no prior art reference or references of record convey or suggest that knowledge, is to fall victim to the insidious effect of a hindsight syndrome wherein that which only the inventor taught is used against its teacher" W.L. Gore & Associates, Inc. v. Garlock Inc., 721 F.2d 1540, 1553, 220 USPQ 303, 312-13 (Fed. Cir. 1983).

It appears that the Examiner has combined the teachings of the prior art with those of the instant application. Only Applicant teaches activation and differentiation of mononuclear cells into predominantly Th1 cells and the subsequent expansion of these cells to a density in excess of 10^{10} cells/liter. Because the combination of cited prior art does not teach or suggest these claim requirements nor provide any motivation to have combined the references, for the rejection to set forth a prima facie case of obviousness, it necessarily must have utilized the teachings of the specification to make the combination. Accordingly, reconsideration and withdrawal of this rejection is respectfully requested.